ON SOME NEW CILIATES LIVING IN THE HIND-GUT OF THE ROACH, *PANESTHIA ANGUSTIPENNIS* ILLIGER.

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WITH 11 FIGURES

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Panesthia angustipennis Illiger is a roach usually found in the rotten wood and sometimes under the bark of more or less decayed part of the trunk of *Pasania* spp., common trees in the southern half of Japan. It lives apparently by devouring dead tissues of the wood. Such a wood-feeding roach is known as a worthy object of study on account of their intestinal protozoa since Cleveland (1934) detected many poly- and hypermastigote flagellates from the hind-gut of an American roach, *Cryptocercus punctulatus*. On examining our *Panesthia angustipennis* Illiger the writer has found no parasitic flagellates but four species of ciliates, one of which seems to be a new species of the genus *Nyctotherus* while three others have shown several unique characters suggesting by themselves the establishment of a new genus. This paper is for recording these four new ciliates.

This work has conveniently been carried on in the laboratory of Otsu Hydrobiological Station as a continuation of the writer's previous studies on the parasitic protozoa and his great obligation is due to Prof. Tamiji Kawamura, the director of the station.

Many individuals of *Panesthia angustipennis* Illiger in various stages of development were found in a forest in a temple yard near the laboratory. The insect were brought to the laboratory with the pieces of rotten wood and kept in a large glass vessel. Provided that the moisture in the vessel is suitable the insects could easily be kept in an apparaently healthy condition. For the examination of the protozoa the intestinal content is placed on a deck glass by means of a smearing method and fixed by Schaudinn's fluid. This is stained by Heidenhain's ironalum haematoxylin and Orange-G, with satisfactory results in all. For the cilial construction Lugol's fluid was applied with a good result.

DESCRIPTION OF THE NEW CILIATES

Family: Plagiotomidae Poche emend. Kahl

Genus: *Nyctotherus* Leidy

1. *Nyctotherus panesthiæ* sp. nov.

This ciliate is abundant in the anterior part of the hind-gut of the host.

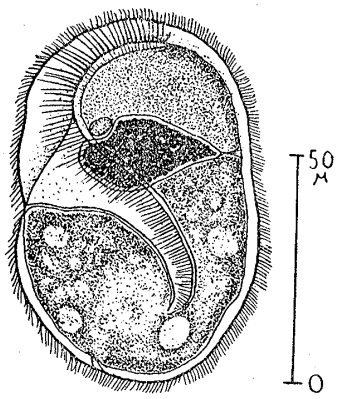

Fig. 1. *Nyctotherus panesthiae.*

The body is kidney-shaped, slightly concave in the ventral side, dark yellow in colour. The change in the shape of the body is slight being not contractile. The body is about 1.5 times as long as wide and is round in the transverse section (Fig. 1). The peristome commences at the anterior tip of the body and continues on the ventral side to the center of the body. The left margin of the peristome is slightly convex and is provided with a row of well developed membranelles, while the right border bears no cilial appendages although developed better than the left. The proximal end of the peristome bends inwards and transits to a well developed cytopharynx. This is a long horn-shaped tube bearing a row of stout cilia on its wall. Running backwards it terminates at a level of from three fourths to four fifths the length of the body.

The thick ectoplasm is clearly marked from the endoplasm. The entire surface of the body is covered densely by fine cilia. They are arranged in numerous meridional rows running from the anterior to the posterior pole of the body. The anal opening can be distinctly observed at the posterior end of the body. The endoplasm is marked off distinctly into two parts. The part anterior to the nucleus shows an alveolar structure and includes neither vacuoles nor food substances, while the other part posterior to the nucleus looks coarsely granular and is provided with many food vacuoles scattered here and there and one, sometimes two, contractile vacuoles near the anal opening.

The macronucleus is large and somewhat discoid in form being thicker in the ventral end than the dorsal end. The micronucleus is a small elliptic body partially embedded in the macronucleus at the anterior corner of the ventral side of the latter. These nuclei are held in the nuclear sac which is stretched horizontally between the wall of

the peristome and the dorsal wall of the body.

The dimensions of the organisms are as follows:

Length of the body.....70–98 μ
Width of the body (in lateral view) .....52–70 μ
Length of the peristome ..... 30 μ
Length of the cytopharynx .....34–46 μ

This form resembles *Nyctotherus reniformis* Bhatia and Gulati in the shape of both the body and cytopharynx and is nearly equal in size. But it deffers distinctly from that in the shape of the macronucleus and presence of the nuclear sac. Some other species may have such a nuclear sac, but they shall easily be distinguished from the present species by either the shape of body or that of cytopharynx.

Genus: *Emmaninius* nov.

The body is spindle-shaped. The anterior part of the body is remarkably produced into a nipple- or neck-shaped portion. In some cases the top end of the body is opened like a funnel. The peristome lies at the anterior end of the produced part and its opening is directed antero-ventrally, or sometimes dorsally owing to a distorsion of the anterior part of the body. The distribution of cilia on the surface of the body is very characteristic, i. e. the body surface is completely naked in the anterior half while densely covered by fine cilia in the posterior half. Many closely arranged cilial striations (groove sculptures in the naked region) are seen on the body surface; they start at about the center of the ventral side of the body and radiate straightly or in a whirl all over the body.

The anal opening is usually distinct but in some cases is comparatively hard to see. It is situated on the ventral surface of the body at the middle point from which the cilial striations start or meet and through which the boundary line between the naked and the ciliated regions of the body passes. The cytopharynx is a well developed tube of long horn-shape, provided with a row of stout cilia on its wall. A large macronucleus and a small micronucleus are enveloped within a nuclear sac which hangs from the dorsal wall of the peristome sustained by a thin fibrous structure. The endoplasm is divided by the nuclear sac into two distinct parts; one, anterior to the nuclear sac, coarsely granular and markedly vacuolated and with one or two contractile vacuoles near the anal opening and the other, posterior to the nuclear sac, alveolar, not vacuolated.

The body is slightly changeable in shape but not contractile. The

peculiar retreating movement is one of the most distinct characteristics of the organisms of the genus.

2. *Emmaninius papilloris* sp. nov.

This species is most abundant in the middle region of the hind-gut of the host. The main part of the body has an inverted ovoid form and its anterior end is protruded to the nipple-shaped peristomal region. The posterior end of the body is thin and conical. The body is slightly compressed laterally and is practically symmetrical in the lateral view. The length of the body is about twice the width in the lateral view (Fig. 2).

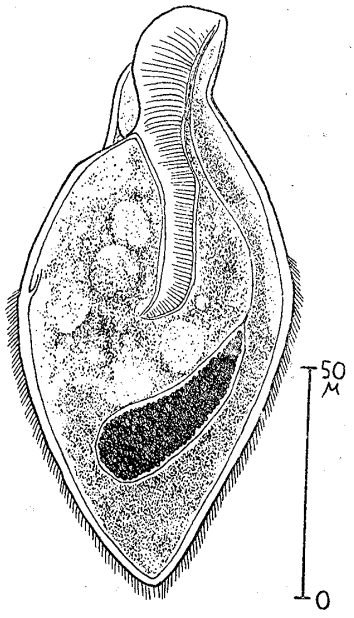

Fig. 2. *Emmaninius papilloris.*

The peristome opens along the antero-ventral side of the nipple-region. The right margin of the peristome is better developed than the left. The latter bears a row of well developed membranelles. The cytopharynx is a long tube; it commences from the proximal end of the peristome and runs backwards along the longitudinal axis of the body. The tip of the cytopharynx reaches the center of the body and its opening is directed ventralwards (Fig. 2).

The ectoplasm is thin but can easily be discriminated from the endoplasm. Both the cilial striations and the groove sculptures start at the center of the ventral side where the anus opens. They diverge radially all over the body surface (Fig. 3). The anal opening is comparatively hard to observe but in some individuals it is detectable at the center of the ventral side of the body (Figs. 2 and 4).

The endoplasm is divided into two regions: one, obliquely anterior to the nucleus, is coarsely granular and highly vacuolated and has a contractile vacuole near the anal opening, the other, posterior to the nucleus, is composed of finely granulated protoplasm. Here is no vacuoles and no food substances and the protoplasm looks rigid (Figs. 2 and 4). The macronucleus is large and situated obliquely in the posterior part of the body. The anterior end of the macronucleus is narrow and pointed and is placed near the center of the dorsal side of the body while its posterior end is thick and broad and is situated near the ventral side at the level about one fifths the length of the body from the posterior end. The micronucleus is a small el-

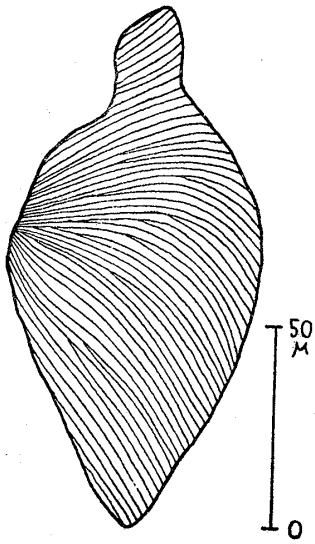

Fig. 3.

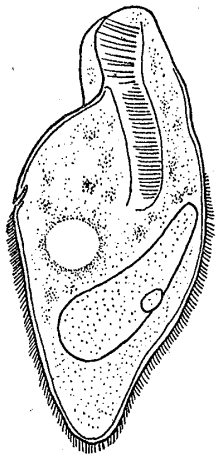

Fig. 4.

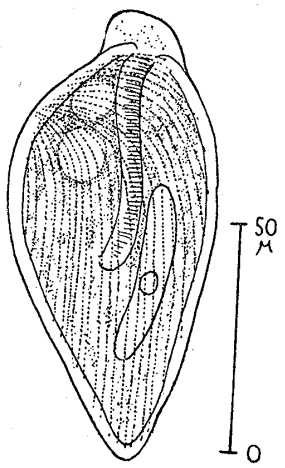

Fig. 5.

Figs. 3 and 4. *E. papilloris.*
Fig. 3. A figure showing cilial striations.
Fig. 4. A figure showing the cytopygium, contractile vacuole and the micronucleus.

E. papilloris in a young stage.

liptic body, situated close to the right side of the macronucleus. The macronuclus is filled by chromatin granules and stained well by hæmatoxylin but the micronucleus is stained only slightly (Figs. 2 and 4). The nuclei are enveloped by a nuclear sac that is hanged by a thin fibrous structure from the dorsal side of the peristome. A part of the finely granulated protoplasm of the posterior end of the body is often produced forewards along the dorso-median line of the body, and the fibrous appendage of the nuclear sac is seen embedded in this protoplasm (Fig. 2).

The dimensions of the organism are as follows:

Length of the body	66–121 μ
Width of the body (lateral)	34—58 μ
Width of the body (ventral)	26—54 μ
Length of the peristome (or the nipple)	12—22 μ
Length of the cytophayrnx	24—40 μ
Length of the macronucleus	24—54 μ
Width of the macronucleus	12—22 μ

The cell division is not observed but many young individuals are found. They are abundant in the anterior half region of the hind-gut of the host (Fig. 5).

The body is ovoid being round at the anterior and narrow at the posterior. The peristome is situated at the anterior end of the body but not yet developed well. The adoral zone is also rudimentary but the cytopharynx is already well constructed. The ectoplasmic

layer is distinct but the anal opening is very hard to detect. The pattern of the cilial striations is similar to that of the main part of the body in the adult individual. The endoplasm is granular and vacuolated but the protoplasmic differentiation is not so remarkable as in the adult state. One contractile vacuole is seen near the anal opening. The nuclei are similar to those of the adult individual but the nuclear sac is not yet developed well.

3. *Emmaninius longicollis* sp. nov.

This species is also found in the anterior half region of the hindgut of the host, but in a very small number. The body is spindle-shaped and is slender than the former species. The anterior part of the body is prominently protruded like a neck and the posterior end is narrow and conical like the former species. The middle portion of the body is thickened and its ventral side is more covex than the dorsal side. The cross section of the body is nearly round. The body length is about 2.5 times the width (Fig. 6).

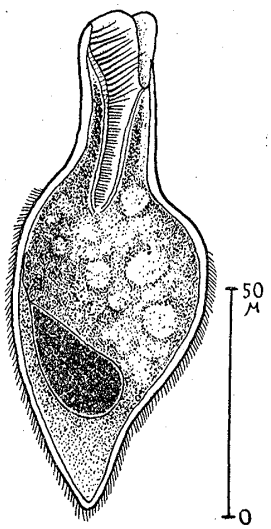

Fig. 6. *Emmaninius longicollis.*

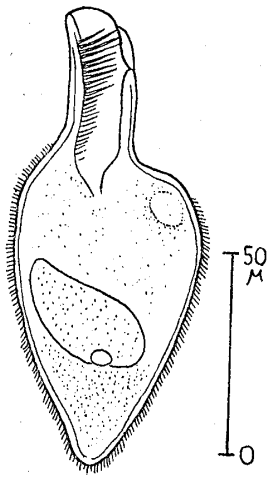

Fig. 7. *E. longicollis* showing the micronucleus.

The peristome commences at the anterior end of the dorsal side of the neck portion and continues nearly till the middle point of the ventral side of the neck. The cytopharynx is well developed it runs straight along the longitudinal axis as far as a distance of about two fifths the body length. As the neck is from one third to two fifths as long as the body, nearly the whole length of the cytopharynx is included in the neck-portion. In this respect the present species differs from *E. papilloris* whose cytopharynx lies wholly in the body region of the organism.

The ectoplasmic cortical layer is distinct. Although the anal opening is almost invisible it must be situated at the center of the ventral side where the cilial striations start in radial lines. The endoplasmic region is divided into two parts as in the case of *E. papilloris*. A single contractile vacuole is seen near the anal opening. The macronucleus

is large and is situated obliquely in the posterior part of the body, the micronucleus is a small round body attaching to the posterior surface of the macronucleus. The nuclear sac is similar to that of the foregoing species (Figs. 6 and 7).

The measurements:

Length of the body	74–110 μ
Width of the body (lateral)	34—42 μ
Length of the neck	24—40 μ
Length of the peristome	10—20 μ
Length of the cytopharynx	20—34 μ
Length of the macronucleus	20—26 μ

This organism closely resembles *E. papilloris* in general organisations but differs distinctly by the slenderness of the body shape, prominently long protruded peristomal region, and the cytopharynx that is located in the neck region.

4. *Emmaninius plantiformis* sp. nov.

This species inhabits exclusively the rectal region of the digestive tube of the host in a very large number. The body is spindle-shaped slightly constricted at the junction between anterior two thirds and posterior one third. The anterior part of the body is protruded, with an aperture of a funnelshape at the tip. The posterior end of the body is conical. The body which is round in cross section is three times as long as the width (Fig. 8).

The opening of the peristome is directed obliquely forewards on the dorsal surface of the body. The right margin of the peristome is better developed than the left. The adoral zone which is composed of a row of long membranelles runs on the left side of the peristome. The peristome bends inwards at its proximal end and connects with the cytopharynx. The latter is a long well developed tube running posteriorly, and reaches to the center of the body. The stout cilia are set on from the wall of the cytopharynx (Fig. 8).

The ectoplasm is thick and distinctly marked from the endoplasm. The boundary line between the naked and ciliated regions of the surface traverses horizontally at the center of the body. The cilial striations as well as the groove sculptures start at the anal opening which is opened at the center of the ventral side as in the cases shown by the former species. But they spread in a whirl, not radially, over the body surface, this is thought to be caused by the distorsion of the anterior part of the body and as the result of which the opening of the

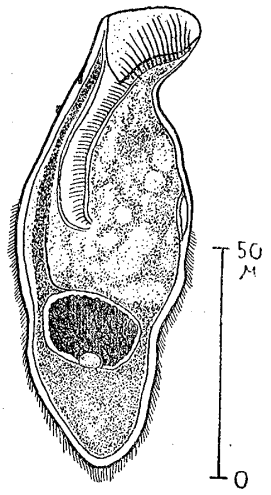

Fig. 8. *Emmaninius plantiformis.*

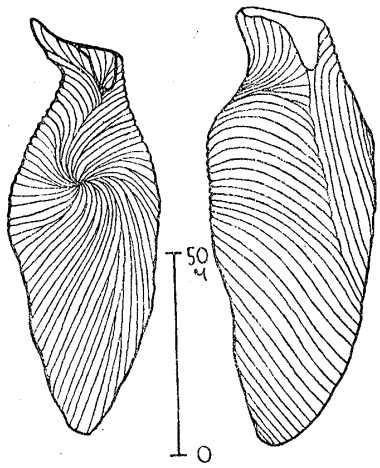

Fig. 9. Fig. 10.
Figs. 9 and 10. *E. plantiforims.* showing cilial striations.
Fig. 9. The ventral surface.
Fig. 10. The dorsal surface.

peristome is also dislocated from the ventral to the dorsal surface of the body (Figs. 9 and 10). The cytopygium is distinct and constantly opened (Fig. 8).

The endoplasm is clearely differentiated into two regions by a constricted portion of the body where the nucleus is located. The protoplasm of the anterior region is coarsely granular and vacuolated while the posterior region is alveolar and looks rigid (Fig. 8). The macronucleus is a thick discoid body and lies horizontally at the constricted part of the body being partially embedded in the alveolar protoplasm of the posterior part of the body. The micronucleus is a small round body situated close to the under surface of the macronucleus. The nuclei are enveloped in a nuclear sac which is suspended by a thin fiber from the dorsal wall of the peristome. A slender extension of the alveolar protoplasm is elongated along the dorso-median line of the body, and the fiber of the nuclear sac runs along this protoplasm.

The measurements of the organism are as follows:

Length of the body	76–132 μ
Width of the body	24—46 μ
Length of the peristome	16—30 μ
Length of the cytopharynx	20—44 μ
Length of the macronucleus (horizontal axis)	16—25 μ
Length of the macronucleus (vertical axis)	10—16 μ

The younger individuals were not scarce (Fig. 11) though no figure of the cell division was observed.

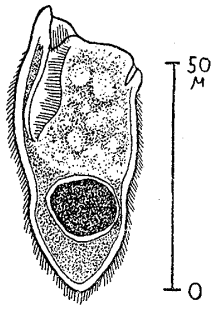

Fig. 11. *E. prantiformis* in a younger stage.

The body is composed only of a ciliated part of

the adult individual. It has a shape like the sole of the human foot. The peristome is situated at a corner corresponding to the first toe of the foot while the anal opening is at the position of the fifth toe. The peristome is not well developed. Only the right side is slightly protruded and the adoral zone is rudimentary. The cytopharynx is developed comparatively well. The body cilia are set on densely on the surface excepting the anterior portion between the peristome and the anal opening. This naked region grows and protrudes in a later stage. The peristomal region is differentiated in the shape of a funnel. The special growth of the peristomal region in the organisms of the present genus seems to be completed in the last stage of the development.

THE SYSTEMATIC POSITION OF *EMMANINIUS* AND THE SIGNIFICANCE OF ITS BODY ORGANISATION

The structure of the adoral zone, the cytopharynx, and the nuclei as well as the degree of the differentiation of the endoplasm suggests the affinities between *Emmaninius* and *Nyctotherus* of the family Plagiotomidæ. *Emmaninius*, however, is characterized by the well developed peristomal region, the dislocated or advanced cytopygium, the peculiar distribution and the striations of the body cilia and the inverse locomotion, and must be separated from any other members of the family. Such coinsidences and distinctions between *Emmaninius* and hitherto known genera of Plagiotomidæ are sufficient diagnoses of this new genus in the family Plagiotomidæ.

The specially well developed peristome of *Emmaninius* is effective when the organism takes food fixing its body to some substratum. Similar cases are seen by the ciliates such as *Stentor* or *Foliculina*. The partial degeneration of the body cilia is by no means uncommon in parasitic ciliates. The remaining cilia of the posterior part of the body are most useful when the organism fixes its body by inserting the posterior part among some substratum in a flow of the intestinal contents in the hind-gut of the host, an inverse locomotion is also effective for this purpose. Similar phenomena are presented by many ciliates who are able to retreat into a specially provided sheath.

Afore mentioned features show that all the species of *Emmaninius* are quite well-adapted to the parasitic life.

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